REMARKS

The present Preliminary Amendment is submitted to delete the multiple dependency of the claims, thereby placing such claims in condition for examination and reducing the required PTO filing fee and to make minor editorial changes so as to generally improve the form of the specification.

Attached hereto is a marked-up version of the changes made to the specification and claims by the current Preliminary Amendment. The attached page is captioned "**Version With Markings to Show Changes Made**".

Respectfully submitted,

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Version with Markings to Show Changes Made

measuring device can discriminate which the correction data is required, only by inserting the biosensor into the measuring device, and there is no need for a user to input the information about the correction data employing a correction chip or the like, thereby removing troubles and preventing operational errors to obtain a correct result.

According to Claim 16 of the present invention, in the biosensor as defined in Claim 15, one or plural fourth slits dividing the electrode part are provided, and the measuring device can discriminate the information of the correction data according to positions of the fourth slits.

Since the biosensor is constructed as described above, the measuring device can discriminate the information of the correction data by the positions of the fourth slits, the correction data can be indicated correspondingly to plural production lots, the measuring device can easily discriminate which correction data is required, by inserting the biosensor into the measuring device, whereby there is no operational trouble, resulting in preventing operational errors to obtain a correct result.

According to Claim [17] 18 of the present invention, in the biosensor as defined in any of Claims 1 to [16] 17, at least one or all of the first slits, the second slits, the third slits, and the fourth slits are formed by processing the electrical conductive layer by a laser.

change occurs between the detecting electrode and the working electrode or the counter electrode of the biosensor, thereby preventing measuring errors due to the shortage of the specimen amount supplied to the reagent layer, resulting in a higher accuracy measurement. Further, when the measurable amount of specimen is supplied to the reagent layer, the measurement is performed by using the detecting electrode also as the counter electrode, thereby making the area of the electrode part smaller, and thus a quantitative analysis based on a slight amount of specimen can be performed correctly.

According to Claim 39 of the present invention, there is provided a quantification method for quantifying, by employing the biosensor as defined in any of Claims 1 to 23 and 37, a substrate included in a sample liquid supplied to the biosensor comprising: a [third] first application step of applying a voltage between the detecting electrode and the counter electrode or the working electrode as well as between the working electrode and the counter electrode; a reagent supplying step of supplying the sample liquid to the reagent layer; a first change detecting step of detecting an electrical change occurring between the [detecting] working electrode and the counter electrode [or the working electrode] by the supply of the sample liquid to the reagent layer; a second change detecting step of detecting an electrical change occurring between the [working] detecting electrode and the counter electrode or the working electrode by the supply of the sample

liquid to the reagent layer; a second application step of applying a voltage between the working electrode and the counter electrode as well as the detecting electrode after the electrical changes are detected in the first change detecting step and the second change detecting step; and a current measuring step of measuring a current generated between the working electrode and the counter electrode as well as the detecting electrode, to which the voltage is applied in the second application step.

Since the quantification is performed as described above, the quantification operation is started when the electrical change occurs between the detecting electrode and the working electrode or the counter electrode of the biosensor, thereby preventing measuring errors due to the shortage of the specimen amount supplied to the reagent layer, resulting in a higher accuracy measurement. Further, when the measurable amount of specimen is supplied to the reagent layer, the measurement is performed by using the detecting electrode also as the counter electrode, thereby making the area of the electrode part smaller, and thus quantitative analysis based on a slight amount of specimen can be performed correctly.

According to Claim 40 of the present invention, in the quantification method as defined in Claim [38 or] 39, the [second] first change detecting step is followed by a no-change informing step of informing a user that no change occurs when it is detected

that no electrical change occurs between the detecting electrode and the counter electrode or the working electrode for a prescribed period of time.

Since the quantification is performed as described above, it is possible to inform a user that there is a shortage of the specimen amount supplied to the reagent layer of the biosensor, resulting in the quantification method with enhanced convenience and safety.

According to Claim 41 of the present invention, there is provided a quantification apparatus, to which the biosensor as defined in any of Claims 1 to 23 and 37 is detachably connected and which quantifies a substrate included in a sample liquid supplied to the biosensor comprising: a first current/voltage conversion circuit for converting a current from the working electrode included in the biosensor into a voltage; a first A/D conversion circuit for digitally converting the voltage from the current/voltage conversion circuit; a first switch provided between the counter electrode included in the biosensor and the ground; and a control part for controlling the fist A/D conversion circuit and the first switch, and the control part applies a voltage between the detecting electrode and the working electrode in a state where the first switch is insulated from the counter electrode, detects an electrical change between the detecting electrode and the working electrode occurring by the sample liquid which is supplied to

the reagent layer on the specimen supply path, thereafter applies a voltage between the working electrode and the counter electrode as well as the detecting electrode in a state where the first switch is connected to the counter electrode, and measures a response current generated by applying the voltage.

Since the quantification apparatus is constructed as described above, measuring errors due to the shortage of the specimen amount supplied to the reagent layer of the specimen supply path are prevented, resulting in a higher accuracy measurement. Further, the detecting electrode of the biosensor is used also as the counter electrode at the measuring, so that the specimen supply path can be downscaled, thereby to perform a quantitative analysis of a slight amount of specimen correctly.

According to Claim 42 of the present invention, there is provided a quantification apparatus, to which the biosensor as defined in any of Claims 1 to 23 and 37 is detachably connected and which quantifies a substrate included in a sample liquid supplied to the biosensor comprising: a [first] second current/voltage conversion circuit for converting a current from the working electrode included in the biosensor into a voltage; a [second] first current/voltage conversion circuit for converting a current from the detecting electrode included in the biosensor into a voltage; a first A/D conversion circuit for digitally converting the voltage from the first current/voltage

conversion circuit; a second A/D conversion circuit for digitally converting the voltage from the second current/voltage conversion circuit; a first selector switch for switching the connection of the detecting electrode of the biosensor to the first current/voltage conversion circuit or the ground; and a control part for controlling the fist first A/D conversion circuit, the second A/D conversion circuit, and the first selector switch, and the control part applies a voltage between the detecting electrode and the counter electrode as well as between the working electrode and the counter electrode in a state where the first selector switch is connected to the first current/voltage conversion circuit, detects an electrical change between the detecting electrode and the working counter electrode as well as an electrical change between the working electrode and the counter electrode, respectively, occurring by the sample liquid which is supplied to the reagent layer provided on the specimen supply path, thereafter connects the first selector switch to the ground, applies a voltage between the working electrode and the counter electrode as well as the detecting electrode, and measures a response current generated by applying the voltage.

Since the quantification apparatus is constructed as described above, measuring errors due to the shortage of the specimen amount supplied to the reagent layer of the specimen supply path are prevented, resulting in a higher accuracy

measurement. Further, the detecting electrode of the biosensor is used also as the counter electrode at the measuring, so that the specimen supply path can be downscaled, thereby to perform a quantitative analysis of a slight amount of specimen correctly.

According to Claim 43 of the present invention, the quantification apparatus as defined in Claim 42 comprises: a second selector switch for switching the connection of the working electrode of the biosensor to the second current/voltage conversion circuit or the ground, and the control part applies a voltage between the detecting electrode and the [counter] working electrode as well as between the working electrode and the counter electrode in a state where the first selector switch is connected to the first current/voltage conversion circuit and the second selector switch is connected to the second current/voltage conversion circuit, respectively, connects the second selector switch to the ground when detecting an electrical change between the working electrode and the counter electrode, occurring by the sample liquid which is supplied to the reagent layer provided on the specimen supply path, and when thereafter detecting an electrical change between the detecting electrode and the working electrode, in a state where the second selector switch is connected to the second current/voltage conversion circuit and the first selector switch is connected to the ground, applies a voltage

between the working electrode and the counter electrode as well as the detecting electrode, and measures a response current generated by applying the voltage.

Since the quantification apparatus is constructed as described above, measuring errors due to the shortage of the specimen amount supplied to the reagent layer of the specimen supply path are prevented, resulting in a higher accuracy measurement. Further, the detecting electrode of the biosensor is used also as the counter electrode at the measuring, so that the specimen supply path can be downscaled, thereby to perform a quantitative analysis of a slight amount of specimen correctly.

According to Claim 44 of the present invention, the quantification apparatus as defined in Claim 42 or 43 comprising an informing means for informing a user that no change occurs, when the sample liquid is supplied to the reagent layer of the specimen supply path, and the control part detects that an electrical change occurs between the working electrode and the counter electrode but no electrical change occurs between the detecting electrode and the working electrode or the counter electrode.

Since the quantification apparatus is constructed as described above, it is possible to inform a user of the shortage of the specimen amount supplied to the reagent layer of the specimen supply path of the biosensor, resulting in the

quantification apparatus with enhanced convenience and safety.

BRIEF DESCRIPTION OF DRAWINGS

Figures 1 are exploded perspective views of a biosensor according to a first and a fifth embodiments.

Figures 2 are diagrams exemplifying how an electrode part is provided.

Figures 3 are exploded perspective views of a biosensor according to a second embodiment.

Figure 4 is a diagram illustrating a specimen supply path of the biosensor according to the second embodiment.

Figure 5 is a top view illustrating a state where slits are formed in an electrical conductive layer of a biosensor according to a third embodiment.

Figures 6 are diagrams illustrating individual wafers of the biosensor according to the third embodiment.

Figure 7 is an exploded perspective view of the biosensor according to the third embodiment.

Figures 8 are diagrams illustrating a state of electrodes of the biosensor according to the third embodiment.

Figures 9 are exploded perspective views of a biosensor according to a fourth embodiment.

Figures 10 are diagrams exemplifying a formation of fourth [second] slits in the biosensor according to the fourth embodiment.

Figure 11 is a schematic diagram showing the concept of a

discriminated according to the electrodes on which the fourth slits which divides the respective electrodes are formed. Therefore, the measuring device can discriminate necessary correction data by inserting the biosensor therein, and thus there is no need for an operator to input correction data by employing a correction chip or the like, resulting in elimination of troubles and a prevention of operational errors. Further, there is provided the reagent layer composed of a reagent which is to be reacted with the sample liquid, the spacer having the cutout part which forms the specimen supply path for supplying the sample liquid to the electrodes, and the cover which is placed on the spacer and has the air hole leading to the specimen supply path, whereby the sample liquid can be easily drawn into the specimen supply path. The electrical conductive layer is formed on the whole surface of the insulating support and is divided into plural electrodes by the first slits, thereby forming high-accuracy electrodes and enhancing the measuring accuracy. Further, since the first slits and the fourth slits are formed by the laser, a high-accuracy processing is possible, whereby the areas of the respective electrodes can be defined with a high accuracy, as well as the clearance between the respective electrodes can be narrowed, thereby to downsize the biosensor.

In any of the above-described biosensors A, B, C, and D according to the first to fourth embodiments, it is more

preferable that each slit provided on the electrical conductive layer is processed by the laser, the width of each slit is 0.005 mm ~ 0.3 mm, and the depth of each slit is equal to or larger than the thickness of the electrical conductive layer, as defined in Claims [16 to 18] (17 to 19) of the present invention.

Further, it is preferred that the reagent layer provided in any of the biosensors A, B, C, and D should include enzyme, an electron transfer agent, or a hydrophilic polymer, as defined in Claims [19 to 21] (20 to 22) of the present invention.

In addition, it is preferable that the insulating support employed in any of the biosensors A, B, C, and D is made of a resin material, as defined in Claim [22] (23) of the present invention.

(Embodiment 5)

A thin film electrode forming method as defined in Claims [23 to 35] (24 to 36) of the present invention will be described as a fifth embodiment with reference to the figures. When the thin film electrode forming method described in the fifth embodiment is applied when the electrode parts of any of the biosensors A, B, C, and D according to the above-described first to fourth embodiments are formed, a biosensor as defined in Claim [36] (37) of the present invention can be obtained.

Figure 11 is a schematic diagram showing a state of a biosensor, where a thin film electrode is formed by implementing the thin film electrode forming method according to this embodiment and a reaction reagent layer are laid out

that value. This indicates that the roughened surface of the support surface reflects the roughened surface of the electrode surface up to the thickness 100 nm, while it reflects the wettability of the electrode material itself (palladium in the embodiment) in the case of the thickness exceeding 100 nm.

Next, the reaction reagent layer including carboxymethyl cellulose as a hydrophilic polymer, glucose oxidase (GOD) as enzyme, and potassium ferricyanide as an electron transfer agent is formed on the thin film electrode which is formed under the above-described conditions, whose thickness of the palladium layer is 10 nm, and thereafter a biosensor for measuring the blood sugar level as in figure 1, in which a spacer and a cover are laid out is manufactured.

Figure 20 is a diagram in which the sensor sensitivities in blood glucose concentrations of 40~600 mg/dl are compared. The blood is drawn into a capillary tube (specimen supply path), then a reaction between a reaction reagent and glucose in the blood is promoted for about 25 seconds, and thereafter a prescribed voltage is applied between terminals of a working electrode and a counter electrode. The sensor sensitivity here is a current value which is obtained 5 seconds after the application of the prescribed voltage. Since the conventional sensor and the sensor in the embodiment have different electrode materials, an applied voltage is 0.5 V for the conventional carbon paste electrode while it is 0.2 V for the palladium thin film

electrode in the embodiment.

Further, the measuring number is n=10 in each concentration range. As apparent from figure 20, it is confirmed that the sensor in the embodiment which is not subjected to a polishing processing or heat processing for the electrode surface has an equivalent or higher sensitivity as compared with a sensor which is subjected to the polishing processing or heat processing, which was conventionally regarded as required to enhance the sensor sensitivity.

The repeatabilities (C.V. values) of the ten-times measuring are compared in (table 1). From the result shown in the table, it is confirmed that the sensor in the embodiment has an excellent accuracy, with variations in individual sensors being reduced, while a conventional sensor has its CV value remarkably deteriorated due to the polishing processing variations or the like.

(Table 1)

Glucose concentration	Conventional sensor	Sensor in embodiment
40mg/dl	15.25%	3.89%
82mg/dl	6.15%	2.87%
165mg/dl	3.89%	2.43%
248mg/dl	3.24%	1.80%
485mg/dl	3.79%	2.16%
600mg/dl	3.28%	1.65%

(Embodiment 6)

Hereinafter, a quantification method of quantifying a

substrate as defined in Claim 38 of the present invention and a quantification apparatus for quantifying a substrate as defined in Claim 41 of the present invention, which employ any of the biosensors A, B, C, and D, for which the electrical conductive layers are formed by employing the above-described thin film electrode forming method according to the fifth embodiment will be described. While the biosensor A as described in the first embodiment is used as a biosensor employed in a following description, the biosensor to be used is not restricted thereto.

Figure 13 is a diagram illustrating structures of the biosensor and the quantification apparatus which is employed in the quantification method employing the biosensor. In the figure, the same reference numerals as those shown in figure 1 denote the same or corresponding parts.

It is a system in which the biosensor A is used in a state where it is connected to a quantification apparatus M1, and the quantification apparatus M1 measures the amount of an included substrate from a specimen supplied to the biosensor A.

In the quantification apparatus M1, numerals 115a, 115b, and 115c denote connectors connected to a working electrode 5, a detecting electrode 7, a counter electrode 6 of the biosensor A, respectively, numeral 116a denotes a switch provided between the connector 115c and the ground (which means a constant potential electrodeposition and can be not always "0". The same goes for in the present specification.), numeral 118a

denotes a current/voltage conversion circuit which is connected to the connector 115a and converts a current flowing between the working electrode 6 and other electrode into a voltage to be output, numeral 119a denotes an A/D conversion circuit which is connected to the current/voltage conversion circuit 118a and converts a voltage value from the current/voltage conversion circuit 118a into a pulse, numeral 120 denotes a CPU which controls ON/OFF of the switch 116a and calculates the amount of a substrate included in a specimen based on the pulse from the A/D conversion circuit 119a, and numeral 121 denotes a LCD (liquid crystal display) which displays a measured value calculated by the CPU 120.

Hereinafter, a description will be given of the operations of the biosensor A and the quantification apparatus M1 when the amount of the substrate included in a specimen is measured by the quantification method employing the biosensor according to the sixth embodiment of the present invention.

First, when the biosensor A is connected to the connectors 115a-115c of the quantification apparatus M1, the switch 116a is turned off under the control of the CPU 120, leading to a non-connection state between the counter electrode 6 and the ground, and a prescribed voltage is applied between the working electrode 5 and the detecting electrode 7. A current generated between the working electrode 5 and the detecting electrode 7 is converted to a voltage by the current/voltage conversion

3. The biosensor as defined in Claim 2, wherein

the counter electrode is provided on the whole or part of the internal surface of the second insulating support,

the working electrode and the detecting electrode are provided on the whole or part of the internal surface of the first insulating support, and

the working electrode and the detecting electrode which are provided on the internal surface of the first insulating support are dividedly formed by the first slits provided on the electrical conductive layer.

4. The biosensor as defined in Claim 1 [or 2], wherein

the electrode part is provided on the whole or part of the internal surface of only the first insulating support, and

the electrode part provided on the internal surface of the first insulating support is dividedly formed by the [fist] first slits provided on the electrical conductive layer.

5. The biosensor as defined in [any of Claims 1 to 4] Claim 1, wherein

an area of the counter electrode is equal to or larger than that of the working electrode.

6. The biosensor as defined in [any of Claims 1 to 4] Claim 2, wherein

a total of an area of the counter electrode and an area of the detecting electrode is equal to or larger than that of the working electrode.

7. The biosensor as defined in Claim 6, wherein

the area of the detecting electrode in the specimen

supply path of the biosensor is equal to the area of the counter electrode.

8. The biosensor as defined in Claim 1 any of Claims 1 to 7, wherein a spacer is provided which has a cutout part for forming the specimen supply path and is placed on the electrode part, and

the second insulating support is placed on the spacer.

9. The biosensor as defined in Claim 8, wherein

the spacer and the second insulating support is integral.

10. The biosensor as defined in Claim 1 any of Claims 1 to 9, wherein an air hole leading to the specimen supply path is formed.

11. The biosensor as defined in Claim 1 any of Claims 1 to 10, wherein the reagent layer is formed by dripping a reagent, and

second slits are provided around a position where the reagent is dripped.

12. The biosensor as defined in Claim 11, wherein

the second slits are arc shaped.

13. The biosensor as defined in Claim 1 any of Claims 1 to 12, wherein third slits are provided for dividing the electrical conductive layer to define an area of the electrode part.

14. The biosensor as define in Claim 13, wherein

shapes of the first insulating support and the second insulating support are approximately rectangular, and

one third slit or two or more third slits are provided in parallel with one side of the approximate rectangle shape.

15. The biosensor as defined in [any of Claims 1 to 14] (Claim 1) having information of correction data generated for each production lot of the biosensor, which correspond to characteristics concerning output of an electrical change resulting from a reaction between the sample liquid and the reagent layer and can be discriminated by a measuring device employing the biosensor.

16. The biosensor as defined in Claim 15, wherein

one or plural fourth slits dividing the electrode part are provided, and

the measuring device can discriminate the information of the correction data according to positions of the fourth slits.

17. The biosensor as defined in [any of Claims 1 to 16] (Claim 1), wherein

at least one or all of the first slits, the second slits, the third slits, and the fourth slits are formed by processing the electrical conductive layer by a laser.

18. The biosensor as defined in Claim 17, wherein

a slit width of respective one of the fist slits, the second slits, the third slits, and the fourth slits is 0.005 mm to 0.3 mm.

19. The biosensor as defined in [Claims 17 and 18] (Claim 17), wherein

a slit depth of respective one of the [fist] (first) slits, the second slits, the third slits, and the fourth slits is equal to or larger than the thickness of the electrical conductive layer.

20. The biosensor as defined in [any of Claims 1 to 19], wherein

the reagent layer includes an enzyme.

21. The biosensor as defined in [any of Claims 1 to 19], wherein

the reagent layer includes an electron transfer agent.

22. The biosensor as defined in [any of Claims 1 to 19], wherein

the reagent layer includes a hydrophilic polymer.

23. The biosensor as defined in [any of Claims 1 to 22], wherein

the insulating support is made of a resin material.

24. A thin film electrode forming method for forming a thin film electrode on a surface of an insulating support including:

a roughened surface forming step of roughening the surface of the insulating support by colliding an excited gas against the surface of the insulating support in a vacuum atmosphere; and

an electrical conductive layer forming step of forming the electrical conductive layer as a thin film electrode which is composed of a conductive substance on the roughened surface of the insulating support.

25. The thin film electrode forming method as defined in Claim 24, wherein

the roughed surface forming step comprises:

a support placing step of placing the insulating support in a vacuum chamber;

an evacuation step of evacuating the vacuum chamber;

a gas filling step of filling up the vacuum chamber with a gas; and

a colliding step of exciting the gas to be ionized and colliding the same against the insulating support.

26. The thin film electrode forming method as defined in Claim 25, wherein

a degree of the vacuum in the evacuation step is within a range of 1×10^{-1} to 3×10^{-3} pascals.

27. The thin film electrode forming method as defined in Claim 26, wherein

the gas is an inert gas.

28. The thin film electrode forming method as defined in Claim 27, wherein

the inert gas is either a rare gas of argon, neon, helium, krypton, and xenon, or nitrogen.

29. The thin film electrode forming method as defined in ~~any~~ claim 24 of Claims 24 to ~~28~~, wherein

the electrical conductive layer forming step comprises:

a second support placing step of placing an insulating support having an already roughened surface, which has been subjected to the roughened surface forming step, in a second vacuum chamber;

a second evacuation step of evacuating the second vacuum chamber;

a second gas filling step of filling up the second vacuum chamber with a second gas; and

a step of exciting the second gas to be ionized and colliding the same against a conductive substance to beat out atoms of the conductive substances, to form a film on the insulating support having the already roughened surface.

30. The thin film electrode forming method as defined in [any] of Claims 24 to [28], wherein

the electrical conductive layer forming step comprises:

a second support placing step of placing an insulating support having an already roughened surface, which has been subjected to the roughened surface forming step, in a second vacuum chamber;

a second evacuation step of evacuating the second vacuum chamber; and

a step of heating and evaporating a conductive substance to deposit steams as a film on the insulating support having the already roughened surface.

31. The thin film electrode forming method as defined in Claim 29 [or 30], wherein

a degree of the vacuum in the second evacuation step is within a range of 1×10^{-1} to 3×10^{-3} pascals.

32. The thin film electrode forming method as defined in [any

of Claims 29 to 31, wherein claim 29

the second gas is an inert gas.

33. The thin film electrode forming method as defined in Claim 32, wherein

the inert gas is either a rare gas of argon, neon, helium, krypton and xenon, or nitrogen.

34. The thin film electrode forming method as defined in any of Claims 29 to 31, wherein claim 29

the vacuum chamber and the second vacuum chamber is the same chamber.

35. The thin film electrode forming method as defined in any of Claims 29 to 34, wherein claim 29

the conductive substance is a noble metal or carbon.

36. The thin film electrode forming method as defined in any of Claims 24 to 35, wherein claim 24

a thickness of a formed thin film electrode is within a range of 3 nm to 100 nm.

37. The biosensor as defined in any of Claims 1 to 23, wherein

the electrical conductive layer is formed by

the thin film electrode forming method as defined in any of Claims 24 to 36.

38. A quantification method for quantifying, by employing the biosensor as defined in any of Claims 1 to 23 and 37, a claim 1 substrate included in a sample liquid supplied to the biosensor

comprising:

a [fist] first application step of applying a voltage between the detecting electrode and the counter electrode or the working electrode;

a sample liquid supplying step of supplying the sample liquid to the reagent layer;

a first change detecting step of detecting an electrical change occurring between the detecting electrode and the counter electrode or the working electrode by the supply of the sample liquid to the reagent layer;

a second application step of applying a voltage between the working electrode and the counter electrode as well as the detecting electrode after the electrical change is detected in the first change detecting step; and

a current measuring step of measuring a current generated between the working electrode and the counter electrode as well as the detecting electrode, to which the voltage is applied in the second application step.

39. A quantification method for quantifying, by employing the biosensor as defined in claim 1 [any of Claims 1 to 23 and 37], a substrate included in a sample liquid supplied to the biosensor comprising:

a [third] first application step of applying a voltage between the detecting electrode and the counter electrode or the working electrode as well as between the working electrode and the

counter electrode;

a sample liquid supplying step of supplying the sample liquid to the reagent layer;

a first change detecting step of detecting an electrical change occurring between the working detecting electrode and the counter electrode or the working electrode by the supply of the sample liquid to the reagent layer;

a second change detecting step of detecting an electrical change occurring between the detecting working electrode and the counter electrode or the working electrode by the supply of the sample liquid to the reagent layer;

a second application step of applying a voltage between the working electrode and the counter electrode as well as the detecting electrode after the electrical changes are detected in the first change detecting step and the second change detecting step; and

a current measuring step of measuring a current generated between the working electrode and the counter electrode as well as the detecting electrode, to which the voltage is applied in the second application step.

40. The quantification method as defined in Claim 38 or 39, wherein

the second first change detecting step is followed by

a no-change informing step of informing a user that no change occurs when it is detected that no electrical change

occurs between the detecting electrode and the counter electrode or the working electrode for a prescribed period of time.

41. A quantification apparatus, to which the biosensor as defined in any of Claims 1 to 23 and 37 is detachably connected and which quantifies a substrate included in a sample liquid supplied to the biosensor comprising:

a first current/voltage conversion circuit for converting a current from the working electrode included in the biosensor into a voltage;

a first A/D conversion circuit for digitally converting the voltage from the current/voltage conversion circuit;

a first switch provided between the counter electrode included in the biosensor and the ground; and

a control part for controlling the first A/D conversion circuit and the first switch,

the control part

applying a voltage between the detecting electrode and the working electrode in a state where the first switch is insulated from the counter electrode,

detecting an electrical change between the detecting electrode and the working electrode occurring by the sample liquid which is supplied to the reagent layer on the specimen supply path,

thereafter applying a voltage between the working

electrode and the counter electrode as well as the detecting electrode in a state where the first switch is connected to the counter electrode, and

measuring a current generated by applying the voltage.

42. A quantification apparatus, to which the biosensor as defined in any of Claims 1 to 23 and 37 (Claim 1) is detachably connected and which quantifies a substrate included in a sample liquid supplied to the biosensor comprising:

a first (Second) current/voltage conversion circuit for converting a current from the working electrode included in the biosensor into a voltage;

a second (first) current/voltage conversion circuit for converting a current from the detecting electrode included in the biosensor into a voltage;

a first A/D conversion circuit for digitally converting the voltage from the first current/voltage conversion circuit;

a second A/D conversion circuit for digitally converting the voltage from the second current/voltage conversion circuit;

a first selector switch for switching the connection of the detecting electrode of the biosensor to the first current/voltage conversion circuit or the ground; and

a control part for controlling the fist (first) A/D conversion circuit, the second A/D conversion circuit, and the first selector switch,

the control part

applying a voltage between the detecting electrode and the counter electrode as well as between the working electrode and the counter electrode in a state where the first selector switch is connected to the first current/voltage conversion circuit,

detecting an electrical change between the detecting electrode and the ~~working~~ counter electrode as well as an electrical change between the working electrode and the counter electrode, respectively, occurring by the sample liquid which is supplied to the reagent layer provided on the specimen supply path,

thereafter connecting the first selector switch to the ground,

applying a voltage between the working electrode and the counter electrode as well as the detecting electrode, and

measuring a current generated by applying the voltage.

43. The quantification apparatus as defined in Claim 42 comprising:

a second selector switch for switching the connection of the working electrode of the biosensor to the second current/voltage conversion circuit or the ground, and

the control part

applying a voltage between the detecting electrode and the ~~counter~~ working electrode as well as between the working electrode and the counter electrode in a state where the first selector switch is connected to the first current/voltage conversion

circuit and the second selector switch is connected to the second current/voltage conversion circuit, respectively,

connecting the second selector switch to the ground when detecting an electrical change between the working electrode and the counter electrode, occurring by the sample liquid which is supplied to the reagent layer provided on the specimen supply path, and

when thereafter detecting an electrical change between the detecting electrode and the working electrode, in a state where the second selector switch is connected to the second current/voltage conversion circuit and the first selector switch is connected to the ground,

applying a voltage between the working electrode and the counter electrode as well as the detecting electrode, and

measuring a current generated by applying the voltage.

44. The quantification apparatus as defined in Claim 42 [or 43] comprising an informing means for informing a user that no change occurs, when the sample liquid is supplied to the reagent layer of the specimen supply path, and the control part detects that an electrical change occurs between the working electrode and the counter electrode but no electrical change occurs between the detecting electrode and the working electrode or the counter electrode.

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IN THE CLAIMS

37. (Amended) The biosensor as defined in [any of Claims 1 to 23] Claim 1, wherein the electrical conductive layer is formed by

[the] a thin film electrode forming method [as defined in any of Claims 24 to 36] including a roughened surface forming step of roughening the surface of the insulating support by colliding an excited gas against the surface of the insulating support in a vacuum atmosphere; and

an electrical conductive layer forming step of forming the electrical conductive layer as a thin film electrode which is composed of a conductive substance on the roughened surface of the insulating support.